

Attorney Docket: 101215-52

IN THE CLAIMS

Amend the claims as follows:

1. (Currently amended) ~~Method~~ A method for the two-dimension ~~dimensional separation of mixtures of biomolecules or other substances mixtures in wherein the biomolecules comprise a solution or an amount of a gel material comprising the biomolecules gels, polymer carriers by means of electrophoresis in an electrophoresis apparatus permitting simultaneous use of two chambers for preparing and performing electrophoresis of two two-dimensional gels, where the method comprising~~

~~the gels casting a first gel for the separation in the first dimension and the gels and casting a second gel for the separation in the second dimension, wherein the first gel and second gel are arranged in succession or simultaneously vertical to one another, wherein casting the first gel and second gel are performed within are brought into an electrophoresis combination chamber, as casting gels or as ready-to-use gels and the first gel is from each other are isolated separated from the second gel by a hollow seal, polymerised and re-hydrated respectively,~~

~~adding buffer solution are then filled in, followed by adding a biomolecule mixture that is deposited onto the gels first gel of the first dimension and the electrophoretic separation of in the first dimension is carried out at constant temperature or at a fixed temperature gradient,~~

~~removing the buffer solution and the hollow seal is then suctioned off, the isolation neutralised and adding a contact gel solution is filled into the resulting spaces between the first gel and second gel dimension and polymerised out, wherein after the contact gel solution polymerizes buffer solutions are filled in and the electrophoretic separation of in the second dimension is carried out at a precisely set temperature and constant electric capacity or increasing current intensity, and~~

~~finally, terminating electrophoresis and developing the gels are developed and to localize the positions of the proteins biomolecules, are made visible by standard methods.~~

2. (Currently amended) Method according to Claim 1, wherein,

Attorney Docket: 101215-52

~~for the separation process~~ first and second gels in the electrophoresis combination chamber are standing vertically and the separation of the proteins is performed in the first dimension vertically and in the second dimension horizontally.

3. (Currently amended) Method according to Claim 1, wherein

~~the gels~~ the first gel of the first dimension ~~are is~~ cast as a flat gels-gel in a U-shaped tube, in which case a stop gel is first cast and, following this, the ~~separation~~ second gel is cast and wherein the gels are cast and polymerized ~~the casting processes as well as the polymerisation processes take place at constant temperature with activated cooling.~~

4. (Currently amended) Method according to Claim 1 wherein

~~the gels of the second dimension are~~ a sealing gel is and cast in ~~two steps in such a way that, in a first step a sealing gel, and after its polymerisation, allowed to polymerize, wherein in a second step the second gel solution is cast from below and rising bottom to top in an upward direction in such a way that the air is displaced upwardly and the gel is finally polymerised at constant temperature with activated cooling.~~

5. (Currently amended) Method according to Claim 1 wherein

the first and second gels are produced with ~~variable~~ different widths and thicknesses.

6. (Canceled)

7. (Canceled)

Attorney Docket: 101215-52

8. (Canceled)

9. (Currently amended) Device for the two-dimensional separation of biomolecules or other substance mixtures in gels by means of electrophoresis in an electrophoresis apparatus indicating comprising electrodes,

wherein

an electrophoresis combination chamber (1) has comprising a core and having two sides (2) with each side comprising cooling elements (3),

the electrophoresis combination chamber further comprising at least one inner gel plate and at least one outer gel plate, wherein the proximity of the inner gel plate and outer plate defines a space for casting gels and buffer reservoirs, and where the each cooling elements makes contact with an inner plate. (3) are arranged between gel chambers (6, 7) and buffer vessels (8) formed on both sides of the core (2) by means of inner plates (4) and outer plates (5) in combination with- and further comprising removable or switchable isolating elements in the form of a hollow seals that support the gels being cast. (9).

10. (Currently amended) Device according to Claim 9,

wherein

the cooling elements (3) are formed by means of a meander-shaped cooling labyrinth (10) with supply (11) and return (12) and the cooling labyrinth (19) encloses the buffer vessels (8) and (21), where the inner plates (4) are made of a good comprise an effective temperature- conducting material and the outer plates (5) are made of a transparent material.

11. (Currently amended) Device according to Claim 9,

wherein

the inner plates (4) consist of ceramic or plastic material and the outer plates (5) of glass or transparent material, and the outer plates (5) are held in position by a clamping frame (13), and a cover (14) that covers off the top of the electrophoresis combination chamber (1) in the upward direction.

Attorney Docket: 101215-52

12. (Currently amended) Device according to Claim 11, wherein

the clamping frame (13) is secured ~~on both sides by~~ means of clamping elements (15) and has viewing windows (16) for process inspection.

13. (Currently amended) Device according to Claim 9,

wherein

~~the lower limitation of the electrophoresis combination chamber (1) is realised by means of an is~~
in a fixed position on an adjustable and rotary table, on which the electrophoresis combination
~~chamber (1) is fixed positioned and the cover (14) indicates comprises~~ inlet and outlet lines for
the cooling medium as well as to the and buffer vessels (8, 21) and gel chambers (6, 7) and the
connections for the electrodes of the first and ~~second dimension.~~

14. (Currently amended) Device according to Claim 9,

wherein

the core (2) ~~consists comprises~~ of a polymer material such as acryl glass, ceramic or plexi-
glass, and the gel chambers (6, 7) are joined with filling tubes (17), and vent openings are
located at the top of each side of the core, arranged, and between inner plates (4) and outer
plates (5) a rectangular seal on each side of the core seals are arranged, and recesses in the
sides of the core for positioning the isolating elements, (9) with recesses act together, and the
~~parts of the electrophoresis combination chamber (1) contacting the media gels and/or gel~~
~~solutions and/or buffer solutions are surface coated, where the surface coating can consist of~~
~~amorphous carbon layers.~~

15. (Currently amended) Device-A device for the two-dimensional separation of mixtures of
biomolecules or other ~~substance substances mixtures~~, wherein the mixture comprises a gel, a

Attorney Docket: 101215-52

~~polymer composition, or a solution, in gels, polymers or carrier free media~~ by means of electrophoresis in an electrophoresis apparatus,

wherein the electrophoresis apparatus comprises

~~all assembly groups as required for the performance of a two-dimensional separation area~~ fully integrated in an electrophoresis combination chamber (1), ~~consisting comprising~~ of a core having two sides, wherein each side comprises ~~(2)~~ with cooling elements (3), wherein the cooling elements make contact with an inner gel plate, the inner gel plate being in proximity of an outer gel plate thereby defining a gel space for casting gels and adding buffer, (3) are arranged between the separating chambers (6,7) formed on both sides of the core by means of inner plates (4) and outer plates (5) in combination and wherein the gel space may be further divided into segments by with removable or switchable isolating-partitioning elements in the form of one or more a hollow seals (9), buffer vessels (8, 21) and holders for the electrodes, and ~~the performance of the two-dimension separation electrophoresis can~~ may be fully automated, ~~without the necessity of manipulation on the gels themselves during the course of the two-dimension separation.~~

16. (Currently amended) Combination chamber for two-dimension separation of mixtures of biomolecules or other substance mixtures by means of electrophoresis in horizontal gels arranged ~~horizontally and~~ above each other ~~by means of electrophoresis with a rear wall plate (28A) and a cover plate (28B), where at least two deflection elements (22) are arranged between rear wall plate (28A) and cover plate (28B) for guiding isolating elements in the form of a hollow seal (24).~~

17. (Currently amended) Combination chamber according to Claim 16,

wherein

the chamber ~~arrangement consists~~ comprises of an upper IEF-part for the performance of the IEF-electrophoresis in the first dimension and a lower part for the performance of the SDS-electrophoresis in the second dimension, ~~and the gels (25) and (26) are arranged horizontally above each other,~~ and the plates (28A, 28B) are sealed off to the outside by means of seals

Attorney Docket: 101215-52

positioned in between the plates, wherein the seals extend along the length of the plates, (23), the configuration of the seals (23) and the thickness of the gels (25, 36) being fixed and, next to the deflection elements (22), electrodes (26, 27) are positioned for the electrophoresis of in the first dimension, and where wherein the rear wall plate (28A) is made of ceramic or glass and the cover plate (28B) is a transparent plate.

18. (Currently amended) Combination chamber according to Claim 16,

wherein

the rear wall plate (28A) forms a unit together with the an upper reservoir comprising a buffer reservoir (29) of for electrophoresis in the second dimension and as well as the a pouring vessel for casting the gel of the second dimension (30), and the a buffer filling vessel for adding a lower reservoir buffer for the second dimension (31), and the an assembly assembled construction consisting comprising of the plates (28A, 28B) and the upper buffer reservoir (29), the pouring vessel (30) and the buffer filling vessel (31) is being positioned placed and arranged in the lower buffer tank (32), and a removable seal positioned between the plates and at the bottom the plates and further, extends the width of the plates, and electrodes in contact with the upper and lower reservoir buffers, (33) is arranged, which is liftable in function, and in the upper buffer reservoir (29) and in the lower buffer tank (32) electrodes (38, 39) are arranged for the electrophoresis of the second dimension.

19. (Currently amended) Method for the two-dimension separation of mixtures of biomolecules or other substance substances mixtures, wherein the mixtures comprise in gels a gel, a or polymer carriers or a solution by means of electrophoresis in an electrophoresis combination chamber, where

- arranging an IEF-gel is arranged horizontally in the combination chamber and is coated over with a re-hydration buffer for re-hydration purposes,
- following this, adding the mixture of a biomolecule biomolecules or other or substance mixture specimen is brought into the IEF-gel or applied to the IEF-gel and the electrophoresis of the first dimension is performed,

Attorney Docket: 101215-52

- ~~before, after or during the performance of the first dimension, casting a horizontal an SDS-gel for the performance, of the second dimension is brought in horizontally to the below and in proximity to the IEF-gel, wherein the SDS-gel and IEF-gel are and, isolated from this separated~~ by a hollow seal, into the combination chamber and the SDS-gel is polymerised,
- ~~removing the hollow seal after completion of the IEF-electrophoresis the isolation is neutralised, and pouring a contact gel put in into the space formerly occupied by the hollow seal, resulting spaces, buffer solution added~~

~~performing the SDS-electrophoresis in the second dimension is carried out, and finally the gels are developed and coloured according to the known methods and~~

~~terminating the electrophoresis in the second dimension, and developing the gel to identify the positions of the biomolecules or other substances.~~

20. (Currently amended) Method according to Claim 19,

wherein

after the re-hydration of the IEF-gel, excess buffer solution is removed and the recess in the IEF-gel is produced by the introduction of a spacer during the re-hydration, and ~~the IEF-gel is re-buffered after the IEF electrophoresis the IEF-gel is contacted with by adding a re-equilibration buffer, and the isolation is neutralised by the the hollow seal is removed removal of a plastic hose by drawing it out with the help of by a stepping motor.~~

21. (Currently amended) Method according to Claim 19,

wherein

~~the cooling gels of both electrophoresis-dimensions are cooled by immersion is performed by immersing the gel sandwiches in thermostatically controlled buffer solution of the second dimension, or alternatively, the buffers and gels are cooled cooling of both electrophoresis-dimensions is realised by cooling chambers by the cooling elements arranged in the core of the combination chamber.~~

Attorney Docket: 101215-52

22. (Currently amended) Method according to Claim 19, wherein

the mixture of biomolecules or other substances ~~biomolecule or substance mixture specimen is~~
brought placed into a recess in the IEF-gel.

23. (Currently amended) Method for the one-dimension separation of mixtures of biomolecules
or other ~~substance substances mixtures~~ by means of electrophoresis, comprising the steps,

- ~~instead of the gel for the separation in the first dimension as well as the isolating~~
~~element, preparing a gel by casting the gel with a gel solution;~~

inserting into the gel solution a comb wherein the comb comprises teeth, each of which
forming a recess or well ~~with specimen pockets for various specimens is placed in the SDS-gel~~
~~and this is polymerised out in the polymerized gel,~~

- removing the comb is removed after out-polymerisation has completed,

- adding an amount of the mixture of biomolecules or other substances ~~the~~
~~specimens are brought into the resulting recesses~~ the recess or well and

- performing electrophoresis following this, the ~~in one-dimension electrophoresis is~~
~~performed.~~

24. (Currently amended) ~~Hydrated~~ A hydrated IEF-gel ~~manufactured prepared~~ by pouring an
immobiline gel solution of with low pK on a gel-polymerised foil and ~~its polymerisation~~ allowing
polymerization to ensue, pouring of an acryl-amide gel solution on the immobiline gel and
~~incipient polymerisation~~ allowing polymerization to ensue, pouring of an immobiline gel solution
of with high pK on the acryl-amide gel and ~~its on polymerisation~~ allow polymerization to ensue,
and subsequent re-hydration ~~by means of with~~ a re-hydration buffer, ~~which contains~~ comprising
1-4% of ~~such~~ ampholines, ~~allowing yielding~~ a pH-range within 2-11.

Attorney Docket: 101215-52

25. (Currently amended) IEF-gel according to Claim 24, wherein

the immobiline gels ~~are manufactured~~ comprise from 6-10% wt-volume of acrylamide and, preferentially 10%, acrylamide with additive of 50-200 mM, preferentially 50-100 mM immobiline.

26. (Currently amended) IEF-gel according to Claim 24, wherein

the acrylamide gel is manufactured from ~~3,5-4,5% 3.5 - 5.0% wt.-vol. of preferentially 3,5-5% acrylamide.~~

27. (Previously presented) IEF-gel according to Claim 24, wherein

the re-hydration buffer contains ~~of 5-9,5 M 5.0 - 9.5M, preferentially 9 M, urea and, as required optionally, detergents, preferentially selected from the group consisting of Tween 20, Chaps or and Triton X-100.~~

28. (Currently amended) IEF-gel according to Claim 24, wherein

before re-hydration, washing steps are carried out as required, and the gel is dried.

29. (Currently amended) A dry Dry-gel manufactured by pouring an immobiline gel solution of with-low pK on a gel polymerisation foil and its-allowing polymerisation, pouring of an acrylamide gel solution on the immobiline gel and its allowing incipient-polymerisation, pouring of an immobiline-containing gel solution of with-high pK on the acrylamide gel and its incipientallowing polymerisation.

30. (New) The IEF-gel according to Claim 26, wherein the acrylamide gel is from 3.5-4.5 wt.-vol% acrylamide.

Attorney Docket: 101215-52

31. (New) The IEF-gel according to Claim 27 wherein the re-hydration buffer contains 9M urea.

32. (New) The method of claim 1, wherein the first gel is a preformed gel that is placed between the inner and outer plates, and rehydrated.